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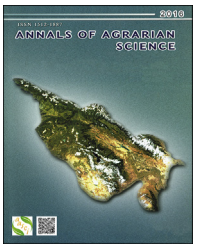

Metabolic control of the insecticides safety use

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ABSTRACT

The results of the conducted research affirm that the phosphororganic insecticides utilization can lead to the break in the nitrogen metabolism, breaking the protein formation, reducing the protein molecules renewal, causing the amino acid and amides accumulation in the active state. It has been revealed that the translocation and transformation of the insecticides under consideration are more closely connected with the changes of insoluble protein fraction. The stagnation point of the Phosphamide and Kaunter impact on the plant has been determined. And only the use of the preparation in optimal norms can influence stimulatingly the course of the process under consideration.

Introduction

Metabolism of a cell, and consequently of the entire plant, is not a simple totality of fermentative reactions divided in space and time but an integral system. It is not possible though to study all the links of this system in a short period of time. That is why we have tried to select the indices, the study of which could reflect both the pattern of metabolism in the most complete way and its connection with the ultimate result, which is the crop capacity and the quality of the wheat grain.

The change in the contents of the nitrogen compositions (proteins, free amino acids, amids, peptides etc.) is of highly importance for wheat. It is connected with the fact that nutritive value and quality of the received flour, as well as colour and taste, in a certain degree are determined by the contents and correlation of the mentioned substances.

It is known that nitrogen metabolism that determines the synthesis of protein substances, significantly affects the proceeding of physiological and bio-chemical processes.

The condition of the plants in the phase of tillering is of great importance as the intense preparation of the plants for wintering takes place during this very phase. The contents of unbound amino acids are an important indicator of the quality of the plants. It is significant because considering quality and contents of unbound amino acids it is possible to get an idea about the condition of the metabolism of nitrogen substances, as well as general physiological condition of the plants. Non-protein fraction of the substances of nitrogen nature, free amino acids being the major part of the latter, is the main reservoir for synthesis and creation of new protein molecules.

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Objectives and methods

In the research aimed at determination of the connection between changes of the wheat metabolism and dynamics of absorption, distribution and accumulation of the phosphor-organic insecticides, there have been used the systemic indices both at the molecular (biochemical processes) and ontogenetic (tolerance of the vegetative organisms at certain ontogenetic stages) levels of the life organization.

The objective of the investigation has been to distinguish the regularities of the impact carried out by various as for their toxicity insecticides transformational processes on the vegetate metabolism in the relations chain «insecticides – plant – crop», considering the reaction of which it would be possible to define the insecticides concentrations dangerous for agricultural ecosystem.

In the research there have been utilized such phosphor-organic preparations as highly toxic Kaunter (Terbufos) and averagely toxic Phosphamide (Dimethoate). In order to distinguish the actions of different factors affecting the vital activity and development of vegetate organisms, there has been used the vegetative method of soil crops. The object of the study is the winter wheat of 80 Poliska sort.

In order to carry out the research the preparations have been brought into the soil taking the culture according to the following scheme: control; different quantities of the insecticides preparations norm (0.25; 0.5; 1.0; 2.0 of the norm), where one insecticide norm is 0.8 kg/ha of the substance in action. The contents of the insecticides under the study in the vegetate material have been defined by the method of fine-layer chronomatography [1]. The determination of the protein contents has been conveyed by the Lawry method [2,3]; the free amino acids have been defined by the gas chronomatography method.

Results and analysis

It is well known that amidst all the metabolism processes it is nitrogen metabolism that influences greatly the proceedings of physiological and biochemical processes identifying the synthesis of protein substances. The contents of unbound amino acids are an important showing of the quality of the plants. It is of highly importance due to the fact that by the quality and contents of unbound amino acids it is possible to get the notion about the condition of the nitrogen metabolism as well as physiological condition of the plants at a whole. Non – protein fraction of the nitrogen substances, the main part of which being made up of unbound amino acids, is the most important reservoir for the synthesis and formation of protein molecules.

The phosphororganic insecticides under the study showed considerable impact on the nitrogen metabolism of winter wheat (Table 1).

The use of just a half of the Dimethoate norm has carried out stimulant effect as for the protein synthesis owing to newly created amino acids and ammonia. The implication of only one norm of this preparation leads to the increase of both protein and non-protein nitrogen contents, and the ammonia accumulated in the plants apparently cannot be completely used for the organic synthesis; this involves intoxication of the plants. Two norms of this preparation inhibit even more significantly the renewal of protein molecules in the vegetate cells which is followed by ammonia accumulation.

The impact of the Kaunter on the nitrogen metabolism has resulted to be even more perceptible. It is the implication of this preparation that shows considerable inhibition of protein synthesis. Only under the application of just one norm of the Kaunter, the metabolism of vegetate organisms turns into more intense organic synthesis with utilization of ammonia and newly grown amino acids.

As a result of Phosphamide and one rate of Kaunter application we can generally observe (Tables 2 and 3) the increase of the contents of irreplaceable heterocyclic amino acids (Histidine, Tryptophan) due to the decrease of the fraction of irreplaceable aliphetic amino acids (Threonine, Leucine, Phenylalanine) (Table 4).

The use of the half and two rates of Kaunter changes the proceedings of the metabolism processes in an opposite way that is especially characteristic of two norms of given insecticide. Knowing the importance of these amino acids in the regulation of vegetate nitrogen balance, it is possible to make an assumption of more intensive protein synthesis in plants with the use of a half and one norm of Phosphamide and one norm of Kaunter. The fact that the products of dissociation of the insecticides under the study take part in biosynthesis of amino acids is proved by the appearance of Cysteine as a result of their utilization, the latter being a kind of amino acid that contains sulphur (Phosphamide contains 29% of sulphur and Kaunter contains 33% of sulphur).

The Lysine contents in the plants has increased almost twice due to the use of two norms of Kaunter; this shows more intensive break in the metabolism in the considered case because Lysine is one of the amino acids that take part in regulation of this process.

As it is generally known, insufficient phosphorus nutrition of the plants leads to the abrupt increase of the contents of the major part of the amino acids, especially Arginine and Proline [4].

Decrease and even absence of Arginine and Proline in all variants of the study apart from the implication of two Kaunter norms, affirms the sufficient phosphorus nutrition of the plants.

It is possible to agree with the authors' opinions, who presume the use of the pesticides to a certain extent as fertilizer. In particular, phosphorganic insecticides have turned out to be peculiar additional source of the phosphorus for the plants of winter wheat. But the application of two norms of Kaunter gives rise to the abrupt increase of Arginine and appearance of Proline. It means that in this case the plants need some additional phosphorus nutrition. The introduction of double norms of phosphorus preparation however could possibly be more intensive phosphorus source for the plants. But here it is apparently found in some bound form, inapproachable for syntactic processes.

More and more adherents are sharing the idea that protein components of protoplastare of primary importance amid the bio-chemical factors of the vegetate defense mechanisms. The ability of a living cell and organism for self-control of

Table 1 – The impact of the phosphororganic insecticides on the ammonia metabolism of thecropof 80 Poliskaw inter wheat sort (the bushying phase).

Operating substance of insecticide, kg/ha		Common maintenance of albuminous nitrogen, %	Common maintenance of amino acid, nanomol, %	NH_3, nanomol, %	Part of ammonia from general nitrogen, %
Control	0.0	–	84.06 ± 0.75	20.19 ± 0.18	24.0
Phosphamide	0.4	112 ± 5.6	51.63 ± 0.46	1.17 ± 0.01	2.3
	0.8	106 ± 5.3	87.30 ± 0.79	29.22 ± 0.26	33.5
	1.6	68 ± 3.4	158.36 ± 1.43	110.50 ± 0.99	69.8
Kaunter	0.2	92 ± 4.6	147.13 ± 1.33	72.70 ± 0.65	49.4
	0.4	96 ± 4.8	73.70 ± 0.66	20.11 ± 0.18	27.3
	0.8	96 ± 4.3	58.02 ± 0.52	15.98 ± 0.14	27.5
	1.6	68 ± 3.4	231.16 ± 2.08	149.42 ± 1.34	64.8

Table 2 – The influence of Phosphamide on the contents of unbound amino acids in the vegetative mass of the 70 Poliska winter wheat sort.

Amino acids	Control	Half of norm	One norm	Two norms
Aspartic acid	0.63 ± 0.01	0.82 ± 0.01	0.83 ± 0.01	0.58 ± 0.01
Threonine	5.97 ± 0.05	4.63 ± 0.04	4.16 ± 0.04	4.19 ± 0.04
Serine	4.45 ± 0.04	4.58 ± 0.04	4.47 ± 0.04	4.75 ± 0.04
Glutamic acid	0.85 ± 0.01	7.43 ± 0.07	5.84 ± 0.05	4.92 ± 0.04
Proline	6.70 ± 0.06	–	–	–
Glycine	1.61 ± 0.01	1.35 ± 0.01	1.50 ± 0.01	1.34 ± 0.01
Alanine	30.68 ± 0.28	36.67 ± 0.33	40.86 ± 0.37	33.30 ± 0.30
Histidine	–	2.55 ± 0.02	2.55 ± 0.02	2.54 ± 0.02
Valine	3.45 ± 0.03	2.92 ± 0.03	2.85 ± 0.03	3.02 ± 0.03
Methionine	1.71 ± 0.02	–	–	1.07 ± 0.01
Isoleucine	2.05 ± 0.02	2.57 ± 0.03	3.12 ± 0.03	3.33 ± 0.03
Leucine	2.82 ± 0.03	–	–	–
Tyrosine	1.26 ± 0.01	3.18 ± 0.03	4.03 ± 0.04	4.27 ± 0.04
Phenylalanine	8.55 ± 0.08	1.20 ± 0.01	–	–
Cysteine	25.81 ± 0.23	31.34 ± 0.28	29.02 ± 0.26	35.89 ± 0.32
Tryptophan	0.39 ± 0.00	0.54 ± 0.01	0.76 ± 0.01	0.81 ± 0.01
Lysine	0.21 ± 0.00	–	–	–
Arginine	2.85 ± 0.03	–	–	–

Table 3 – The influence of Kaunter on the contents of unbound amino acids in the vegetative mass of the 70 Poliska winter wheat sort.

Amino acids	Control	Half of norm	One norm	Two norms
Aspartic	0.63 ± 0.01	–	1.00 ± 0.01	–
Threonine	5.97 ± 0.05	3.12 ± 0.03	3.86 ± 0.04	5.81 ± 0.05
Serine	4.45 ± 0.04	2.39 ± 0.02	4.04 ± 0.04	4.12 ± 0.04
Glutamic	0.85 ± 0.01	2.91 ± 0.03	5.33 ± 0.05	0.83 ± 0.01
Proline	6.70 ± 0.06	–	–	2.08 ± 0.02
Glycine	1.61 ± 0.01	0.56 ± 0.01	1.22 ± 0.01	1.74 ± 0.02
Alanine	30.68 ± 0.28	38.29 ± 0.35	39.48 ± 0.36	38.80 ± 0.35
Histidine	–	–	3.01 ± 0.03	–
Valine	3.45 ± 0.03	0.14 ± 0.00	4.61 ± 0.04	3.09 ± 0.03
Methionine	1.71 ± 0.02	–	–	1.06 ± 0.01
Isoleucine	2.05 ± 0.02	1.36 ± 0.01	3.57 ± 0.03	1.97 ± 0.02
Leucine	2.82 ± 0.03	0.89 ± 0.01	–	3.46 ± 0.03
Tyrosine	1.26 ± 0.01	–	5.36 ± 0.05	1.37 ± 0.01
Phenylalanine	8.55 ± 0.03	6.94 ± 0.06	–	4.46 ± 0.04
Cysteine	25.81 ± 0.23	39.94 ± 0.36	26.56 ± 0.24	25.37 ± 0.23
Tryptophan	0.39 ± 0.00	–	1.05 ± 0.01	–
Lysine	0.21 ± 0.00	0.11 ± 0.00	–	0.38 ± 0.00
Arginine	2.85 ± 0.03	1.87 ± 0.02	–	5.48 ± 0.05

Table 4 – Dynamics of fractional composition of proteins winter wheat plants under the influence of organophosphorus insecticides, % of control.

Variant		Shoots				Tillering	
		First leaf		Third leaf			
		Soluble proteins	Insoluble proteins	Soluble proteins	Insoluble proteins	Soluble proteins	Insoluble proteins
Phosphamide	0.5 of norm	100 ± 1.5	88 ± 1.8	98 ± 1.4	65 ± 2.1	98 ± 1.3	155 ± 2.3
	1 norm	92 ± 1.4	89 ± 1.9	97 ± 1.5	50 ± 1.8	108 ± 1.0	100 ± 2.3
	2 norms	101 ± 1.6	61 ± 1.1	103 ± 1.3	59 ± 2.0	74 ± 1.3	50 ± 2.2
Kaunter	0.25 of norm	125 ± 1.8	68 ± 1.3	149 ± 1.4	40 ± 0.5	96 ± 1.4	81 ± 0.6
	0.5 of norm	110 ± 1.3	87 ± 1.8	141 ± 1.4	50 ± 1.1	96 ± 1.3	94 ± 0.6
	1 norm	66 ± 1.3	104 ± 1.8	125 ± 1.3	69 ± 1.0	93 ± 1.3	104 ± 0.7
	2 norms	147 ± 1.1	19 ± 1.2	128 ± 1.3	43 ± 0.5	74 ± 1.8	50 ± 1.0

proper metabolism, for its automatic reorganization in the optimal direction with the change of inner and external conditions, as well as for the quick realization of the adaptation reactions, are determined first of all by the proteins as "flexibility and movement of the protein molecule conformation provides its capacity to adequately respond to the evidence of certain factors in the environment" [5]. But the utilization of proteins usually takes place in several stages. Firstly, their mobilization takes place which is followed by conformation reconstruction for realization of polypeptide bonds hydrolysis. Consequently, such properties of proteins as solubility are changed. Then, immediate protein breakdown into primary peptides and amino acids which constitute them begins [6,7]. That is why we have been studying in our work the impact of the insecticides under investigation on the changes in the vegetative mass of the winter wheat different according to for the protein fractions dissolution: soluble proteins (movable proteins of the cell structures) and insoluble proteins (structural proteins of chloroplasts, mitochondria, cytoplasmic reticulum).

More intensive accumulation of soluble proteins fractions under the influence of Kaunter is observed (Fig. 1b), this can be connected with the acceleration of the proteins renewal. One norm of use has appeared an exception, as here in the primary period of the insecticide impact we can observe considerable (34%) oppression of the soluble proteins fractions. In the further, more flowing transformation which leads to the increase of this fractionis observed in the variant of Phosphamide and abrupt increase in the variant of Kaunter. It is necessary to mention that Phosphamide has revealed less impact on the soluble proteins fractions than Kauter has, because the changes of the contents of this fraction under the influence of Phosphamide on the winter wheat proceed more flowingly than under the influence of Kaunter.

Concerning insoluble proteins fractions, the oppression due to the impact of both preparations is observed. Contrary to the expecting it has turned out that translocation and transformation of the insecticides under the investigation are more closely connected with the changes of this very fraction (Fig. 1a and b).

The increase of the insoluble proteins fractions happens in direct proportion to the accumulation of the insecticides in the plants before concentration which coincides with the "susceptibility point", this can be considered the critical point of the impact of this insecticide on the plant. The concentration of the preparation in the plant above this «point» causes sudden decrease of the fraction.

In the phase of tillering after the complete plant detoxication of the insecticides we can observe the following: the use of 0.5 norm of Phosphamide led to the sudden 55% increase of the insoluble proteins content without changing the soluble fraction. The use of one norm of Phosphamide has caused merely noticeable impact on the fractional contents of the protein (the 8% increase of the soluble fraction); two

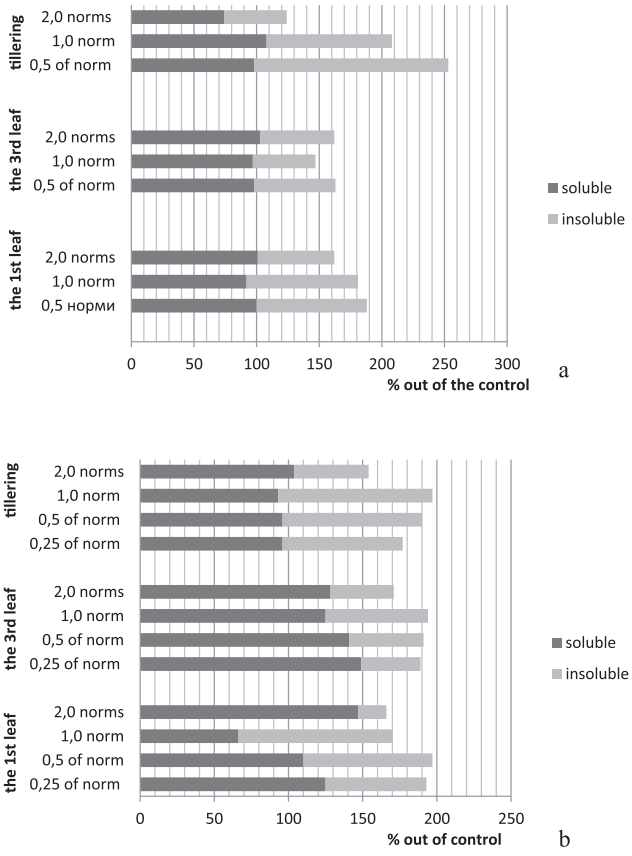

Fig. 1 – The dynamics of the fractional protein contents of the winter wheat under the influence of phosphorganic insecticides (a – Phosphamide, b – Kaunter).

norms of this preparation have abruptly reduced the contents of both fractions, especially the insoluble one (the 50% decrease). As for the Kaunter, the use of only one norm has led to the inconsiderable (4%) increase of the insoluble protein fraction. The impact of other concentrations of this insecticide on the winter wheat has caused considerable oppression of both protein fractions; this has been especially noticeable (25% of soluble and 50% insoluble protein fractions) when using two norms of the preparation.

Conclusion

It has been revealed that the use of phosphorganic insecticides can lead to the breakdown of the protein metabolism, impeding the protein formation and reducing the renewal of protein molecules.

We have noticed the direct dependence between the toxicity of the preparation and the degree of its impact on the metabolic vegetative processes.

It has also been determined that the use of the preparations in the optimal concentrations can cause the stimulating influence on the proceeding of the nitrogen metabolism (in our case it is 0.5 of norm of Phosphamide which is 0.4 kg/ha of operating substance and one norm of Kaunter which is 0.8 kg/ha of operating substance).

Thus, in order to reveal environmentally harmful concentrations of pesticides with the example of phosphorganic insecticides, it is possible to suggest the study of the nitrogen metabolism of the plants under the influence of these substances.

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